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| HEALTHY HOMES INITIATIVE (HHI) BACKGROUND INFORMATION External Review Draft, Version 2 October 2, 2001 | | | |
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Healthy Homes Issues: Mold

External Review Draft, Version 2 October 2, 2001

Prepared for:

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Preface

In October 1998, in response to Executive Order 13045 on "Protection of Children from Environmental Risks and Safety Risks," the U.S. Department of Housing and Urban Development (HUD) launched the Healthy Homes Initiative (HHI). The primary goal of the HHI is to protect children from housing conditions that are responsible for multiple diseases and injuries. As part of this initiative, HUD is preparing a series of papers to provide background information to their current HHI grantees, as well as other programs considering adopting a healthy homes approach. This background paper focuses on molds, and provides a brief overview of the current status of knowledge on:

- The extent and nature of mold hazards in the home:
- Assessment methods for mold hazards in the home;
- Mitigation methods for mold hazards in the home; and
- Information needs in the field of mold research

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Table of Contents

| 1.0 | OVERVIEW OF THE PROBLEM | | |
|------|---|---|-----|
| 2.0 | EXTENT AND NATURE OF MOLD HAZARDS IN THE HOME | | |
| | 2.1 | Environmental and Housing Factors Affecting Mold Growth | 1 |
| | 2.2 | Exposure and Health Effects | |
| | | Allergens | |
| | | Toxics and Irritants | |
| 3.0 | MET | HODS USED TO ASSESS MOLD HAZARDS IN THE HOME | 4 |
| | 3.1 | Visual Assessment | 5 |
| | 3.2 | Sample Collection | 5 |
| | | Source Sampling | 6 |
| | | Air Sampling | 7 |
| | 3.3 | Sample Analysis | 8 |
| | | Culture Methods and Spore Examination | 8 |
| | | Chemical Analyses | 9 |
| | | Immunoassays | 10 |
| | | Genetic probes | 10 |
| | 3.4 | Interpretation of Results | 11 |
| 4.0 | MET | HODS USED TO MITIGATE MOLD HAZARDS IN THE HOME | 13 |
| | 4.1 | Guidelines for Mitigation and Personal Protection | 13 |
| | | Moisture Control | 15 |
| | | Removal and Cleaning of Mold Contaminated Materials | 15 |
| 5.0 | CUR | RENT RESEARCH AND INFORMATION GAPS | 16 |
| Refe | ences | | 18 |
| Anne | ndix A. | Additional Internet Resources | A-1 |

Healthy Homes Issues: Mold

1.0 OVERVIEW OF THE PROBLEM

There are over 200 species of fungi to which people are routinely exposed indoors and outdoors (NAS, 2000). This includes mold-like fungi, as well as other fungi such as yeasts (unicellular fungi forming pasty colonies) and mushrooms, which are characterized by the familiar fruiting bodies people think of as "mushrooms." The terms "mold" and "mildew" are non-technical names commonly used to refer to any fungus that is growing in the indoor environment (Burge and Otten, 1999). These names are used interchangeably, although mildew is often applied to growths on fabrics, window sills or bathroom tiles. Because molds and mildews may be any of several natural classes of fungi, these names are not interchangeable with the nomenclature used in biological classification systems (Burge and Otten, 1999).

In general, molds are characterized by a vegetative body composed of a network (mycelium) of threadlike filaments (hyphae), which infiltrate the mold's food or habitat. Mold colonies may appear cottony, velvety, granular, or leathery, and may be white, gray, black, brown, yellow, greenish, or other colors (Burge and Otten, 1999). Many reproduce via the production and dispersion of spores. They are usually saprophytes (i.e., they feed on dead organic matter) and, provided with sufficient moisture, can live off of many materials found in homes, such as wood, cellulose in the paper backing on drywall, insulation, wallpaper, glues used to bond carpet to its backing, and everyday dust and dirt.

Research indicates that certain molds can cause a variety of adverse human health effects, including allergic reactions and immune responses (e.g., asthma), infectious disease (e.g., histoplasmosis¹), and toxic effects (e.g., aflatoxin-induced liver cancer) (ACGIH, 1999). Molds are thought to play a role in asthma in several ways. They are known to produce a large number of compounds that are potentially allergenic, and there is sufficient evidence to support associations between fungal allergen exposure and asthma exacerbation and upper respiratory disease (NAS, 2000). In addition, molds may play a role in asthma via release of irritants that increase potential for sensitization, or release of toxins (mycotoxins) that affect immune response (NAS, 2000). Finally, mold toxins can cause direct lung damage leading to pulmonary diseases other than asthma (NAS, 2000).

2.0 EXTENT AND NATURE OF MOLD HAZARDS IN THE HOME

2.1 Environmental and Housing Factors Affecting Mold Growth

In indoor environments, mold originates from two sources: mold infiltrating from outdoors (e.g., through open windows), and mold colonization on the interior of the home. Molds can obtain nutrients and moisture sufficient for growth from water-affected building materials such as wallboard and insulation materials, as well as carpets, furniture, and clothing. Using a score system based on material bioavailability, Gravesen et al. (1999) evaluated the susceptibility of

EXTERNAL REVIEW DRAFT

¹ A disease caused by the inhalation of spores of the fungus *Histoplasma capsulatum* (associated with bird or bat droppings); disease is most often asymptomatic but occasionally produces acute pneumonia or an influenzalike illness and spreading to other organs and systems in the body.

various building materials to mold attack. They found that the products most vulnerable to mold attack were water damaged, aged organic materials containing cellulose, such as wooden materials, jute, wallpaper, and cardboard.

Different fungal species vary with regard to environmental conditions required for optimal growth, but all are influenced by moisture, temperature, light, and the substrate nutrient concentration and type (Burge and Otten, 1999). One of the most important factors affecting mold growth in homes, however, is moisture level. In general, most molds require fairly wet conditions (near saturation), lasting for many days, to extensively colonize an environment (NAS, 2000). In addition to affecting the extent of mold colonization, moisture availability also affects the types of mold present. For example, certain *Penicillium* species grow in relatively dry environments (e.g., in house dust with a high relative humidity), while others, such as Stachybotrys species, require continuously wet substrates such as soaked wallboard, water reservoirs for humidifiers, or drip pans (Burge and Otten, 1999; Bush and Portnoy, 2001). Relative humidity also affects spore release for some molds (e.g., Aspergillus and Penicillium), with spore release occurring with lowering humidity after initial growth at high humidity levels (Foarde et al., 1997a). Furthermore, as moisture availability changes, it has been observed that the species composition (i.e., the rank order of dominant species) will also often change. Some of the most abundant fungi genera found in homes without severe water damage include: Alternaria, Cladosporium, Penicillium, yeasts, and Aspergillus (Burge and Otten, 1999; American Academy of Pediatrics, 1998; Bush and Portnoy, 2001; Gravesen et al., 1999). Most of these molds do not typically produce mycotoxins (Etzel, 2000), but may be important as sources of mold allergens. In contrast, under certain very damp conditions (i.e., in the presence of water-soaked cellulosic materials), toxin producing Stachybotrys chartarum may be prominent (Flannigan, 1997). In general, whether or not a potentially toxigenic fungi produces toxins is dependent on environmental conditions and nutrient source (Burge and Ammann, 1999).

Housing features that can increase moisture levels and growth of mold include poor ventilation, excess production or condensation of water in the house (e.g., humidifiers, unvented clothes dryers), and water leakage or flooding (Lawton et al., 1998; Gravesen et al., 1999). Li and Kendrick (1995, as cited in Bush and Portnoy, 2001) found that overall fungal levels (as assessed by counting spores in environmental samples) were highest in living rooms, followed by family rooms, kitchens, bathrooms, and bedrooms. Also in this study, it was observed that fungal levels increased with the presence of damp conditions and carpets, and decreased where forced-air heating systems, dehumidifiers, air filters, and air conditioners were present. Douwes et al. (1999) also found that fungal levels, as assessed by measurement of extracellular polysaccharide (EPS) fungal cell wall components from *Aspergillus* and *Penicillium* species (EPS-*Asp/Pen*), were highest in living room floor dust. In addition, EPS-*Asp/Pen* levels were 2 to 3 times higher on carpeted floors than on smooth floors.

2.2 Exposure and Health Effects

Mold exposure in homes primarily occurs via inhalation of airborne spores and hyphal fragments. Molds are also present in household dust and on surfaces, with exposure occurring when particles are disturbed and become airborne or, less commonly in residential situations,

through dermal contact or ingestion. Release of mold spores or fragments into indoor air from mold colonies is usually dependent on some sort of mechanical disturbance, although for some types of molds slight air movement may be sufficient (e.g., air movement by a fan), or spores may become airborne through natural spore discharge mechanisms. Most molds release spores ranging in size from 2 to 10 μ m, although some may be released as chains or clumps of spores (NAS, 2000).

Allergens. Many molds produce numerous protein or glycoprotein allergens capable of causing allergic reactions in people. These allergens have been measured in spores, as well as other fungal fragments. An estimated 6-10% of the general population and 15-50% of those who are genetically susceptible (atopic) are sensitized to mold allergens (NAS, 2000). Some of the major mold allergens identified and isolated to date include those from *Aspergillus fumigatus*, *Aspergillus oryzae*, *Alternaria alternata*, *Cladosporium herbarum*, *Penicillium citrinum*, *Penicillium chrysogenum*, *Trichophyton tonsurans*, *Malassezia furfur*, and *Psilocybe cubensis* (NAS, 2000). Research clearly indicates that exposure to mold plays a role in the exacerbation of asthma symptoms in sensitized individuals, although the association between mold exposure and asthma development remains undetermined (NAS, 2000). The clearest association between mold exposure and asthma is for sensitization to *Alternaria* (generally regarded as an outdoor mold), although this may be because the allergens of this genus (*Alt a* 1 and *Alt a* 2) are well characterized relative to other mold species (NAS, 2000; Platts-Mills and Woodfolk, 2000). Information on the nature of exposures that lead to mold-related asthma is lacking (ACGIH, 1999; NAS, 2000).

Toxics and Irritants. Many molds are also known to produce toxic metabolites (mycotoxins) that can be a health hazard upon ingestion, dermal contact, or inhalation. An overview of some common molds, mycotoxins, and associated health effects is presented in the American Conference of Government of Industrial Hygienists publication *Bioaerosols: Assessment and* Control (ACGIH, 1999). While common outdoor molds present in ambient air, such as Cladosporium cladosporioides and Alternaria alternata, do not usually produce toxins, many other different mold species do (Burge and Ammann, 1999). Genera producing fungi associated with wet buildings, such as Aspergillus versicolor, Fusarium verticillioides, Penicillium aiurantiorisen, and Stachybotrys chartarum, can produce potent toxins, measurable in mold mycelia, spores, and the matrix in which the mold is growing (Burge and Ammann, 1999). A single mold species may produce several different toxins, and a given mycotoxin may be produced by more than one species of fungi. Furthermore, a toxin-producing fungi does not necessarily produce mycotoxins under all growth conditions, with production being dependent on the substrate it is metabolizing, temperature, water content and humidity (Burge and Ammann, 1999). Because species of toxin-producing molds generally have a higher water requirement than common household molds, they tend to thrive only under conditions of chronic and sever water damage (American Academy of Pediatrics, 1998). For example, Stachybotrys typically only grows under continuously wet conditions (Burge and Otten, 1999).

Although epidemiological studies that specifically examine exposure to mycotoxins in indoor residential environments are relatively limited, there is substantial evidence of a relationship between mycotoxin exposure (via ingestion and inhalation) and adverse health effects in occupational (agricultural and food processing) settings and animal studies (Rao et al., 1996;

Miller, 1994; American Academy of Pediatrics, 1998; Burge and Ammann, 1999). The most frequently studied mycotoxins are produced by species of Aspergillus (e.g., aflatoxins), Fusarium, Penicillium, Stachybotrys, and Myrothecium (e.g., satratoxins, trichothecenes) (Burge and Ammann, 1999). Known health effects depend on the kind of mycotoxin and the nature of the exposure, but include mucous membrane irritation, skin rashes, dizziness, nausea, immunosuppression, and birth defects (Burge and Ammann, 1999). Although evidence is very limited in residential environments, aflatoxins (produced by Aspergillus flavus and parasiticus), have also been linked to liver cancer in food processing settings (Burge and Ammann, 1999). Toxins from Stachybotrys chartarum have been most commonly associated with lung inflammation and hemorrhage in animal studies (Nikulin et al., 1996, 1997, as cited in Burge and Ammann, 1999) and non-specific symptoms (headaches, sore throats, flu symptoms, diarrhea, fatigue, and dermatitis) in case studies (Dill et al., 1997 and Croft et al., 1986, as cited in Burge and Ammann, 1999). In indoor environments, associations have also been reported for pulmonary hemorrhage deaths in infants and the presence of Stachybotrys chartarum (Etzel et al., 1998; Flappan et al., 1999; Elidenir et al., 1999; Vesper et al., 2000). Although this specific association has not been conclusive (CDC, 2000), recent research does clearly support the potential for general mycotoxin exposure in the indoor environment to result in adverse effects on respiratory health (NAS, 2000; Sorenson, 1999, Rao et al., 1996; American Academy of Pediatrics, 1998). It has also been suggested that very young children may be especially vulnerable to certain mycotoxins (American Academy of Pediatrics, 1998; Etzel, 2000). For example, Etzel (2000) suggests that exposure to the trichothecene mycotoxins, which are known to be potent protein synthesis inhibitors, may result in pulmonary capillary fragility in the rapidly growing lungs of children younger than one year.

Other mold associated compounds, including glucans and volatile organic compounds (VOCs), are also suspected to play a role in certain adverse reactions described as "sick building" or "building related symptoms" (Burge and Otten, 1999). Glucans are a major component of the cell walls of most molds, and have been observed to have irritant effects similar to (but less potent than) those of bacterial endotoxins. VOCs, which are produced by molds as byproducts of growth or degradation of substrates, may also be responsible for some non-specific building related symptoms; however, the role of VOCs in specific disease has not been studied (Burge and Otten, 1999)

3.0 METHODS USED TO ASSESS MOLD HAZARDS IN THE HOME

In general, visual observation of active or past microbial growth or measurement of mold in dust or samples of source material can be used to establish potential for mold exposure. As inhalation is the primary exposure pathway for molds, air sampling for mold can be used to estimate the likelihood of exposure (Dillon et al., 1999).

The following section provides the reader with an overview of the range of assessment methods and technologies that are available, from both a research and programmatic perspective. The level of rigor involved in assessing mold hazards in a research setting generally surpasses that which is practical or necessary for programmatic or public health use. From a housing or public health perspective, a home assessment is generally constrained by the need for cost

October 2, 2001

effective methods that are sufficient to allow for the identification of molds which may be at levels of concern in the home environment.

3.1 Visual Assessment

High humidity levels and excess dampness have clearly been associated with mold growth, as well as several other home health hazards, such as dust mites. Visual inspection for dampness, observable mold growth, and detection of musty odors, often obtained from occupant questionnaires, are the most frequently used methods to assess the potential for indoor mold exposure. Visual observation of mold growth, however, is limited by the fact that fungi are microscopic and their presence is often not apparent until growth is extensive. In addition, destructive sampling (e.g., the removal of wallboard) is often required to assess the extent of fungal contamination (Dillon et al., 1999). A device called a boroscope, which employs fiber optics technology to make observation in wall cavities and behind wall board through a small hole drilled in the wall board, can be used by home inspectors to facilitate assessment of hidden mold damage in a fairly non-destructive manner (Greenberg, pers. comm.). Although direct observation of visible fungal growth is usually sufficient to warrant a recommendation for mitigation, further air or source sampling (discussed below) may be conducted for documentation purposes and to record the types of fungi that predominate (Burge and Otten, 1999).

Many moisture problems in homes are due to structural deficiencies. Common points of inspection for buildings with dampness problems include: rain leaks (e.g., on roofs and wall joints); surface and groundwater leaks (e.g., poorly designed or clogged rain gutters and footing drains, basement design problems); plumbing leaks; and stagnant water in appliances (e.g., dehumidifiers, dishwashers, refrigerator drip pans, and condensing coils and drip pans in HVAC systems). In addition, assessment is also conducted for water vapor migration and condensation problems, including: uneven indoor temperatures, poor air circulation, air conditioning systems, soil air entry into basements, contact of humid unconditioned air with cooled interior surfaces, and poor insulation on indoor chilled surfaces (e.g., chilled water lines). Portable, hand-held moisture meters may also be useful in qualitative home assessments to aid in pinpointing areas of potential biological growth that may not be otherwise obvious during a visual inspection (ACGIH, 1999). A variety of different protocols exist for assessing water damage in homes; for example, a visual assessment tool for inspecting homes for evidence of mold and moisture has been developed for Cleveland, Ohio, by the Cuyahoga County Board of Health (Dillon et al, 1999; Allan, pers. comm.). An overview of additional techniques and issues of concern in conducting visual assessments of homes for mold contamination is presented in *Bioaerosols*: Assessment and Control (ACGIH, 1999; see Chapter 4, "The Building Walkthrough").

3.2 <u>Sample Collection</u>

Quantitative assessment of indoor molds generally involves sampling of a representative environmental medium in the home and quantification of the measure of interest (e.g., allergen level, total fungal biomass, or spore count). Because preparation requirements for environmental samples vary with the analysis techniques to be used, investigators should plan a collection procedure accordingly. Air and dust sampling, as well as direct sampling of mold

colonies or bulk materials where visible mold growth is present, are commonly used to estimate environmental levels and hazard potential for molds. However, the relationship between mold levels in these media and actual exposure is unknown. Generally, indoor environments contain large reservoirs of mold spores and hyphal fragments in settled dust and contaminated building materials, of which only a relatively small amount is airborne at a given time. Standard methods for quantitative sampling of mold, or models that would allow for estimates of inhalation or dermal exposure to molds from sampling results, are not available (Dillon et al., 1999).

Source Sampling. Source sampling methods used in investigations of mold contamination in homes includes bulk and surface sampling.

In bulk sampling techniques, portions of environmental materials (e.g., settled dust, sections of wallboard, pieces of duct lining, carpet segments, or return air filters) are collected and tested to determine if molds have colonized a material and are actively growing, and to identify surfaces areas where previously airborne mold spores and fragments have settled and accumulated (Martyny et al., 1999). For fixed materials, bulk samples are cut or otherwise removed from the source and thus this technique may be somewhat destructive. For loose materials, such as floor dust, bulk samples are typically collected using wipe sampling or a hand-held vacuum with a special filter. Various factors, including design of the vacuum collector, surface characteristics (e.g., carpet vs. smooth floor), and other environmental characteristics have all been shown to affect the efficiency of dust collection (Wang et al., 1995; NAS, 2000). For example, Wang et al. (1995) observed that when collecting dust with a vacuum sampler from a shag carpet surface, lower relative humidity (e.g., around 20 percent, as would be encountered during a dry, cold season) increased the intensity of the electrostatic field on the carpet and thus significantly decreased the collection efficiency of the vacuum. Standardized methods for collecting household dust samples have been developed by researchers studying lead and pesticide exposures, as those used, for example, in HUD's National Survey of Lead and Allergens in Housing (Clickner et al., 2001). In the National Survey, single wipe dust samples for lead analysis were collected by the technique described in ASTM E 1728-95, with one sample taken from the center of the largest open area of each selected room. These and other reports containing dust sampling methods are available on HUD's website at http://www.hud.gov/offices/lead/.

Surface sampling in mold contamination investigations may also be used when a less destructive technique than bulk sampling is desired. For example, non-destructive samples of mold may be collected using a simple swab or adhesive tape. In general, surface sampling is typically accomplished by either pressing a collection material (e.g., a contact plate or adhesive tape) against a surface, or by wiping an area with a wetted swab, cloth, or filter (Martyny et al., 1999). The size of a collected surface sample is generally much smaller that that of a bulk sample. An overview of procedures and advantages of various contact sampling techniques, including agar plate methods, adhesive tape sampling, and surface-wash sampling, is presented in *Bioaerosols: Assessment and Control* (ACGIH, 1999; see Chapter 12, "Source Sampling" by Martyny et al., 1999).

Air Sampling. For routine assessments in which the goal is to identify possible mold contamination problems prior to remediation, it is usually unnecessary to conduct air sampling because decisions about appropriate remediation strategies can typically be made on the basis of a visual inspection (NYC, 2000). Air monitoring may, however, be necessary in certain situations, including: 1) if an individual has been diagnosed with a disease associated with fungal exposure, 2) if it is suspected that the ventilation systems are contaminated, or 3) if the presence of mold is suspected but cannot be identified by a visual inspection or bulk sampling (NYC, 2000).

Airborne mold particulates may include spores, fungal fragments, aggregates of spores or fragments, or materials contaminated with fungal product. The most commonly used methods available today for volumetric air sampling are based on one of the following principles: inertial compaction (e.g., multiple-hole impactors, slit samplers), centrifugal collection (e.g., agar-strip impactors, cyclone samplers), filtration (e.g., cassette filters attached to portable pumps), and liquid impingement (e.g., three-stage impingers) (Martyny et al., 1999). Gravitation or settling techniques (e.g., longer-term collection of settled spores onto a culture plate or microscope slide) can also be used, but due to large temporal and spatial variations, gravity techniques cannot be used as a substitute for volumetric measurements (O'Meara and Tovey, 2000; Martyny et al., 1999).

Samplers may be either static or reflective of a personal breathing zone. Static filter samplers used to collect airborne substances are normally placed in a fixed position in a room and do not measure personal exposure (O'Meara and Tovey, 2000). Sampler design and flow rate have been shown to affect the quantity and size of particles sampled, and thus can affect the apparent measured levels of a given airborne substance (O'Meara and Tovey, 2000). Both high-volume (60 to 1100 L/min) and lower-volume (6 to 20 L/min) filter samplers have been used, although it has been suggested that the lower-volume samplers may collect a more meaningful sample in relation to exposure because they better approximate breathing volumes of humans (O'Meara and Tovey, 2000). Breathing zone samplers often show much higher levels of collected mold particles than static samplers, likely due to the varying levels of dust that are resuspended in the personal breathing zone as a result of human activity; however, only minor differences in airborne mold levels between personal and static samplers are observed during high levels of dust disturbance (O'Meara and Tovey, 2000). It is generally recommended in the literature that outdoor air samples are collected concurrent with indoor samples for comparison purposes, both for measurement of baseline ambient air conditions (remote from obvious mold sources), and for baseline measurement of air entering a building (samples near outdoor air intakes) (ACGIH, 1999).

An overview of the principles of airborne sample collection and several commercially available air sampling instruments is provided in *Bioaerosols: Assessment and Control* (ACGIH, 1999; see Chapter 11, "Air Sampling" by Willeke and Macher). Factors for consideration when selecting a particular type of air sampler for fungal collection are also discussed in Chapter 5 of the ACGIH publication. For example, it is recommended that consideration be given to such factors as: the compatibility of the sampler with the analysis method to be used, what type of information is needed (e.g., concentration or identification of species), the concentration (e.g., very high or very low) of the mold at the test site, temperature extremes, the nature of the air

stream where the sample will be collected, and possible collection constraints due to the presence of occupants (ACGIH, 1999). Comparative assessments of the performance of the different samplers (e.g., filter samplers, Andersen samplers, rotorod samplers, liquid impingers, and cyclone samplers) have been inconclusive, although certain samplers have been observed to perform better for specific purposes (e.g., the Andersen six-stage sampler for viable spore counts and the Burkard 24-hour samplers for total spore counts) (Flannigan, 1997). Many factors introduce significant variability into air sampling results and complicate interpretation as discussed in Section 3.4.

3.3 <u>Sample Analysis</u>

Current methods available to analyze environmental samples from the home for mold hazards include:

- Counting colonies cultured for specific species
- Identifying and/or counting spores
- Chemical analysis of fungal components to quantify total fungal loads (biomass)
- Immunoassays (ELISAs) to measure allergen levels
- Genetic probe technologies to identify fungal species

No single method provides a complete assessment of the exposure hazard associated with an environmental sample, as discussed in Section 3.4 below. The quality of environmental microbiology laboratories performing analyses on samples for molds and other microbiological agents is monitored under an external peer review program sponsored by the American Industrial Hygiene Association (AIHA). This program, which includes the Environmental Microbiology Proficiency Analytical Testing (EMPAT) Program (a performance evaluation program) and the Environmental Microbiology Laboratory Accreditation Program (EMLAP), is specifically for labs identifying microorganisms commonly detected in air, fluids, and bulk samples during indoor air quality studies. When a laboratory is accredited by AIHA, the laboratory and its clients have the assurance that the laboratory has met defined standards for performance based on examination of a variety of criteria. Proficiency in EMPAT is mandatory for labs seeking EMLAP accreditation. Additional information on the EMPAT and EMLAP programs is available online at http://www.aiha.org/micro.html.

Culture Methods and Spore Examination. The growth of fungal colonies on specially prepared nutrient media (culture) from spores contained in air or dust samples is a common method used to assess mold populations. Following culture, identification of fungal species can often be accomplished with a dissecting or light microscope via examination of colony morphology or spore bearing structures. Culture results can also be reported in terms of colony forming units (CFU) per m³, g, or cm². The type of isolation media used to culture the fungal spores, however, can introduce substantial variability into the types and relative magnitudes of mold species that are cultured (Burge and Otten, 1999; Flannigan, 1997). Bias in culture measurements may be introduced because a highly nutritionally rich substrate favors the growth of fast-growing species, or because one species present in the sample may not compete well with another on the culture plate (Flannigan, 1997). For example, some genera such as *Penicillium* grow well and quickly on most media and thus may be over-represented in a

culture sample, while others such as *Stachybotrys* grow slowly or not at all on commonly used substrates (Bush and Portnoy, 2001).

Many types of fungi are identifiable (at least to general category) via microscopic examination of spores in collected air and source samples (Burge and Otten, 1999). Spores counts can also be reported, typically in units of spores per m³, g, or cm².

Chemical Analyses. Methods using chemical analysis can be used to quantify total fungal loads (biomass), although, generally, these methods do not allow for identification of species. These methods can be based on chemical components (biomarkers) common to a particular group of organisms (e.g., ergosterol in the membranes of fungal hyphae and spores), or on other fungal components which are directly associated with health effects (i.e., β -glucan in cell walls of hyphae and spores) (Flannigan, 1997). Results of dust analysis are typically expressed as concentration in units of weight of analyte per weight of settled dust (e.g., ng/g for allergens and toxins, μ g/g for glucans or ergosterol). Results of air sample analysis are usually expressed volumetrically.

Mycotoxins. Methods currently available for detecting mycotoxins in environmental samples were designed for testing agricultural products and generally do not translate well to residential testing requirements (e.g., air samples with very low mycotoxin concentrations) (Burge and Ammann, 1999). Thin layer chromatography has been used to measure mycotoxins in some studies, although the usefulness of this technique is limited due to lack of sensitivity and susceptibility to interference (Burge and Ammann, 1999). High performance liquid chromatography (HPLC) and gas chromatography with mass spectrometric detection (GC-MS) have also been used for mycotoxin quantitation, although these techniques are also limited due to specialized laboratory requirements and associated expense (Burge and Ammann, 1999). Various researchers have measured cell toxicity of particulate air samples and inferred the presence of mycotoxins. For example, Vesper et al. (2000) used a protein synthesis inhibition assay to evaluate the toxicity of air particulate samples during a Stachybotrys chartarum remediation study. Protein synthesis inhibition is an activity characteristic of trichothecene mycotoxins typically produced by *Stachybotrys*. Field sample extracts were assayed for trichothecene toxicity by comparison to a known sample, with the results expressed as toxin equivalents per cubic meter of air. Mycotoxin analysis can be used to detect the presence of certain fungi in the environment, but, more commonly, mycotoxin levels are only measured after the fungal species has been identified (Bush and Portnoy, 2001).

Other Chemical Components of Fungi. Ergosterol, which is a component of fungal cell membranes, has been used as an index of fungal mass in house dust and air samples, and can be analyzed using gas chromatography with mass spectrometric detection (Flannigan, 1997; Dillon et al., 1999). Ergosterol is not present in vascular plants, and therefore, in most indoor environments can be used as a specific measure of fungal mass (Dillon et al., 1999). Ergosterol measurement has been applied in assessments of house dust and air (Dillon et al., 1999), although, as with mycotoxin analysis, this highly specialized technique may have resource limitations for home assessments

β (1 \rightarrow 3)-D-glucan, a component of fungal cell walls, can be analyzed using a modification of the *Limulus amoebacyte* lysate (LAL) technique or enzyme inhibition assay (EIA), and has also been used to measure total fungal biomass in house dust and air (Dillon et al., 1999; Flannigan, 1997). However, β (1 \rightarrow 3)-D-glucan is not specific to fungal cell walls and may originate from plants and some bacteria (Douwes et al., 1998). In addition, a standardized protocol for the storage, extraction, and analysis of environmental samples for β-glucan is not well developed (Dillon et al., 1999).

Mold extracellular polysaccharides (EPS) have potential usefulness as fungal measures, as they are produced in mycelial cell walls under almost all growth conditions (Dillon et al., 1999). Douwes et al. (1999) examined the relationship between measured EPS from *Aspergillus* and *Penicillium* species (EPS-*Asp/Pen*) and culturable fungi, reported home dampness, and respiratory symptoms. EPS-*Asp/Pen* levels were significantly correlated with total culturable fungi, and levels in living room floor dust were positively associated with home dampness and respiratory symptoms. EPS can be measured using a specific enzyme inhibition assay (EIA), although the determination of EPS has not yet been routinely applied in indoor studies (Dillon et al., 1999).

There are about 15 volatile organic compounds (VOCs) produced by fungi that may also be used as markers of fungal growth, although some are also emitted by bacteria (Dillon et al., 1999). VOCs can be collected on solid sorbents, extracted, and quantified using gas chromatography with mass spectrometric detection. Measurement of fungal VOCs may be particularly useful in some home assessments for detection of hidden mold growth because the compounds can permeate porous walls in buildings (Dillon et al., 1999). However, the uncertainties currently associated with accuracy of these methods preclude using this approach for routine investigations. For example, significant questions remain regarding reliable "signature" VOCs for a particular fungus, and how to deal with the variability in VOC produced under different conditions (Ammann, 1999).

Immunoassays. To measure mold allergen levels in collected dust and air samples, enzymelinked immunosorbent assays (ELISAs, or also commonly called immunoassays) have been developed for numerous indoor mold allergens. Immunoassays are a laboratory technique that make use of the specific binding between the antigen associated with an allergen and its homologous antibody in order to identify and quantify a substance in a sample. However, although immunoassays have been developed for many major fungal allergens to date, this technology is not as highly developed or well-standardized as that for house dust mite, cat, or cockroach allergens (Bush and Portnoy, 2001). Only assays for *Alternaria* (Alt a 1) and *Aspergillus* (Asp f 1) are currently widely available from commercial laboratories (e.g., see Indoor Biotechnologies website at http://www/inbio.com/index.html) (Vailes et al., 2001).

Genetic probes. Polymerase chain reaction (PCR) based technologies (i.e., genetic probes), unlike other non-culture methods, can be used to identify certain biological particles such as fungi to the species level (Flannigan, 1997). The technology is based on targeting short, species-specific sequences of DNA. EPA's Office of Research and Development, National Exposure Research Laboratory, has recently been refining a DNA-based system that allows rapid identification and quantification of molds in a matter of hours. Although a technique not

yet widely available, at least one commercial lab (Aerotech, Inc. in Arizona ²) is offering analysis of indoor samples (preferably dust, but can be applied to any medium) using genetic probes (Vesper, personal communication). Genetic probes have not yet been developed for identifying allergen proteins specifically (Vesper, personal communication). These methods could prove particularly useful in situations where fungi are not otherwise easily differentiated on the basis of morphology (e.g., *Aspergillus* and *Penicillium*) or where culture methods are not useful because spores have lost their viability (O'Meara and Tovey, 2000).

3.4 <u>Interpretation of Results</u>

Methods for assessing human exposure to fungal allergens and mycotoxins are relatively poorly developed (NAS, 2000) and interpretation of results is difficult. This is due, in part, to the fact that fungal allergens and toxins vary widely across mold species, and because the traditional methods of mold population assessment (e.g., spore counts) do not have consistent relationships with levels of mold allergens or toxins. Furthermore, because viable mold measures do not include particles that are not culturable (non-viable spores or non-reproducing vegetative fragments) but that may have toxic or allergenic properties, investigations of moldaffected houses that focus only on assessing the number of culturable organisms may underestimate actual allergenic or toxic potential (Flannigan and Miller, 1994; Flannigan, 1997). Conversely, total measures of a fungal component (e.g., ergosterol or glucan) in a sample do not allow for identification of mold species, or provide information about the biologically active portion of the sample. Therefore, neither measure provides a complete assessment of the potential allergen or mycotoxin exposure hazard associated with an environmental sample. The accuracy of substituting measures of exposure to fungi for exposure to fungal allergens or toxins has not been determined (ACGIH, 1999), and direct measurement of allergens and toxins is limited by the current development and standardization of immunoassays for specific allergens and reliable, affordable techniques for mycotoxin analysis.

Further complicating the exposure assessment is variability associated with the collection of samples. The accuracy of quantifying air samples is complicated by large variations in airborne concentrations from room to room and temporally over relatively short periods of time, as well as outdoor concentrations with season (O'Meara and Tovey, 2000; Flannigan, 1997; Flannigan and Miller, 1994). Dust sampling for molds is sometimes used to circumvent this temporal variability, although dust samples sometimes show differences in the relative abundance and types of mold in comparison to air samples (Flannigan, 1997; Dillon et al., 1999). The release of molds from carpets and walls or other surfaces has also been cited as an important factor in introducing variability into the magnitude and nature of indoor air spora collected (Flannigan, 1997). In addition, due to the ubiquitous presence of mold spores in the outdoor environment (often in concentrations far higher than indoors), it can be difficult to establish the presence of indoor mold growth using air sampling.

Finally, there is the issue of comparison of results to standards that indicate potential hazard. The major limitations with existing quantitative guidelines for fungi are the lack of human

² Aerotech Laboratories, Inc. Phoenix, Arizona. Additional information is available by calling (800) 651-4802 or on the internet at www.aerotechlabs.com/index.htm#

dose/response data, reliance on short term grab samples analyzed only by culture methods, and the lack of standardized protocols for data collection, analysis, and interpretation (Rao et al., 1996). For example, Verhoeff and Burge (1997) conducted a review of peer-reviewed literature through 1995, and identified nine population based studies that examined the relationship between allergy and the presence of fungi in the home environment. All of the studies included quantitative measures of fungal presence in either air or dust. Evaluation of the studies indicated that although the existence of positive associations between fungal levels and health outcomes was supported in the literature at that time, inconsistency and inadequate validation of the measures used to evaluate exposure and health effects made determination of guidelines for fungi in home environments based on health risk assessment impossible (Verhoeff and Burge, 1997).

Currently, there are no standard numerical guidelines for assessing whether there is a mold contamination problem in an area. In the U.S., there are no EPA regulations or standards for airborne mold contaminants (USEPA, 2001). Various governmental and private organizations have, however, proposed guidance on the interpretation of fungal measures of environmental media in indoor environments (quantitative limits for fungal concentrations). These organizations include the ACGIH, the U.S. Occupational Safety & Health Organization (OSHA), the American Industrial Hygiene Association (AIHA), the Canada Mortgage and Housing Corporation (CMHC), the Commission of the European Communities (CEC), and the World Health Organization (WHO), as well as numerous smaller and/or local organizations like the New York Department of Health. The only government agency that had binding quantitative regulations for airborne fungi is the Russian Federation (Rao et a., 1996). Reviews of guidance offered by various groups to assist investigators in the interpretation are available in Bioaerosols: Assessment and Control (ACGIH, 1999) and in Rao et al (1996). Recommendations reported in Rao et al (1996) vary widely, with quantitative standards/ guidelines ranging from less than 100 CFU per m³ to greater than 1000 CFU per m³ as the upper limit for airborne fungi in non-contaminated indoor environments (Rao et al., 1996). Bush and Portnoy (2001) suggest that indoor spore counts equal to or greater than 1000/m³ and colony counts on the order of 1000 to 10,000 CFU per m³ likely represent indoor fungal contamination.

Other factors in addition to indoor spore counts may also be considered. For example, the University of Minnesota, Department of Environmental Health and Safety recommends consideration of several factors in addition to total spore counts when attempting to assess the severity of a mold contamination problem, including: the number of fungi indoors relative to outdoors, whether the fungi are allergenic or toxic, if the area is likely to be disturbed, whether there is or was a source of water or high relative humidity, if people are occupying the contaminated area or have contact with air from the location, and, whether there are immune compromised individuals or individuals with elevated sensitivity to molds in the area (University of Minnesota, 1996).

Given evidence that young children may be especially vulnerable to certain mycotoxins (American Academy of Pediatrics, 1998), and in view of the potential severity or diseases associated with mycotoxin exposure, some organizations support a more conservative approach to limiting mold exposure (Burge and Otten, 1999). For example, the American Academy of

Pediatrics recommends that infants under 1 year of age are not exposed at all to chronically moldy, water-damaged environments (American Academy of Pediatrics, 1998).

4.0 METHODS USED TO MITIGATE MOLD HAZARDS IN THE HOME

4.1 Guidelines for Mitigation and Personal Protection

Common intervention methods reported in the literature for residential mitigation of mold hazards include:

- Location and removal of sources of moisture (control of dampness and humidity and repair of water leakage problems),
- Increasing ventilation,
- Cleaning of mold contaminated materials,
- Physical removal of materials with severe mold growth,
- Use of high-efficiency air filters,
- Maintenance of heating, ventilation, and air conditioning systems, and
- Prevention of spore infiltration from outdoors by closing doors and windows and by using air conditioning.

The literature also consistently emphasizes the importance of worker protection when conducting mold assessment and mitigation projects. Activities such as cleaning or removal of mold-contaminated materials in homes, as well as investigations of mold contamination extent, have the potential to disturb areas of mold growth and release fungal spores and fragments into the air. For example, Vesper et al. (2000) measured a very high number of *Stachybotrys* spores in personal breathing zone samples of a worker during the implementation of a mold remediation program to remove *Stachybotrys* contaminated materials (i.e., wallboard, paneling and carpeting) from water damaged areas of a home. This suggested that residents should not attempt repairs without the proper protection, or preferably should employ a contractor trained in environmental remediation (Vesper et al., 2000). Recommended measures to protect workers during mold remediation efforts depend on the severity and nature of the mold contamination being addressed, but include the use of well fitted particulate masks or respirators that retain particles as small as 1 µm or less, disposable gloves and coveralls, and protective eyewear (ACGIH, 1999).

Various guidance documents for remediation of mold contamination have been developed.

- The New York City Department of Health has a set of guidelines, "Assessment and Remediation of Fungi in Indoor Environments," originally developed for *Stachybotrys* but expanded to be inclusive of all molds, that are widely recognized (available at http://www.nyc.gov/html/doh/html/epi/moldrpt1.html).
- The Institute of Inspection Cleaning and Restoration Certification produced guideline S500: Standard and Reference Guide for Professional Water Damage

Restoration (available by contacting the IICRC headquarters at (360) 693-5675 or through e-mail at supplies@iicrc.org).

- The American Conference of Governmental Industrial Hygienists (ACGIH) bioaerosols committee published in 1999, "Biosaerosols: Assessment and Control," a compilation of information on investigation strategies, sampling and analysis, and control of indoor bioaerosols, including molds (can be ordered from ACGIH at http://www.acgih.org/home.htm).
- The American Industrial Hygiene Association (AIHA) is in the process of developing a document with explicit guidelines for mitigation of mold hazards and some general guidelines for "clearance".
- U.S. Environmental Protection Agency published guidance for "Mold Remediation in Schools and Commercial Buildings," which includes many general principles also applicable to residential mold mitigation efforts (available from EPA online at http://www.epa.gov/iaq/molds/index.html).
- The Canada Mortgage and Housing Corporation published, "Clean-up Procedures for Mold in Houses," which provides qualitative guidance for mold mitigation (can be ordered from CMHC at http://www.cmhc-schl.gc.ca/boutique/en/)
- Health Canada published its "Fungal Contamination in Public Buildings" guide to assist investigators in recognizing and managing fungal contamination (available from Health Canada online at http://www.hcsc.gc.ca/ehp/ehd/catalogue/bch.htm#technical).

Although these and other mold remediation guidance documents share many of the same approaches for conducting residential mitigation of mold hazards, such as correction of moisture problems and removal of severely contaminated materials, specific criteria cited in the guidelines may vary. For example, ACGIH (1999) guidance regarding remediation techniques and personal protective equipment (PPE) is based on qualitative professional judgment of the extent of fungal contamination (defined as minimal, moderate, or extensive), while USEPA (2001) guidance for mold remediation in schools is based on quantitative estimates of the total surface area affected (defined as small (less than 10 ft²), medium (between 10 and 100 ft²), or large (greater than 100 ft²)). The New York City guidelines (NYC, 2000) differentiate between large isolated areas of contamination (30 to 100 ft²) and extensive contamination (greater than 100 contiguous ft² in an area).

In general, however, the literature agrees on the point that a particular strategy or combination of strategies recommended for a given mold abatement effort (including the degree of worker protection needed) will depend on site-specific factors, such as the contaminating agent, the type of substrate that is contaminated (e.g., whether porous or non-porous), the extent of the contamination, the location of the site requiring remediation, and the presence of highly susceptible occupants (ACGIH, 1999; Morey, 2000). For example, slight fungal contamination

of a semi-porous concrete floor may only require cleaning, while extensive mold growth in a carpet will require complete removal. Appropriate PPE and containment measures for situations of minimal colonization (small isolated surface area contamination) might include contaminated source containment to minimize dust or spore dispersion (e.g., dust suppression methods such as misting, covering material with sticky sheeting or an encapsulant prior to removal) and the use of a N-95 disposable respirator and gloves for PPE (ACGIH, 1999; NYC, 2000). For moderate contamination or where sensitive individuals are present in the home, containment of the source by enclosing the work area with a plastic sheet and sealing with tape and negative pressurization may be warranted (NYC, 2000; ACGIH, 1999). In many cases, the protection and mitigation methods most appropriate must be determined using professional judgment, and it is often recommended that investigators seek additional advice, when needed, from occupational physicians, toxicologists, respiratory protection experts, or health and safety professionals to select appropriate PPE (USEPA, 2001; ACGIH, 1999).

For many indoor contaminants, integrated approaches for indoor environmental interventions are considered most effective. For integrated abatement of fungal contamination, guidance sources generally recommended that abatement strategies take into account both the control of new sources of exposure, and the removal or cleaning of reservoirs. In the control of new sources, it is also emphasized that the infiltration of spores from outside, as well as the growth of fungi indoors, needs to be considered (Bush and Portnoy, 2001; Eggleston and Bush, 2001).

Moisture Control. Because one of the most important factors affecting mold growth in homes is moisture level, controlling this factor is crucial in mold abatement strategies. Many simple measures can significantly control moisture, for example: maintaining indoor relative humidity at no greater than 50%-60% through the use of dehumidifiers, fixing water leakage problems, increasing ventilation in kitchens and bathrooms by using exhaust fans, venting clothes dryers to the outside, reducing the number of indoor plants, using air conditioning at times of high outdoor humidity, heating all rooms in the winter and adding heating to outside wall closets, and using a sump pump in basements prone to flooding (Bush and Portnoy, 2001; ACGIH, 1999; NYC, 2000).

Removal and Cleaning of Mold Contaminated Materials. Non-porous (e.g., metals, glass, and hard plastics) and semi-porous (e.g., wood and concrete) materials contaminated with mold and that are still structurally sound can often be cleaned with detergent and bleach solutions or by using quaternary amine preparations; however, in some cases, the material may not be easily cleaned or may be so severely contaminated that it may have to be removed. It is recommended that porous materials (e.g., ceiling tiles, wallboards, and fabrics) that cannot be cleaned be removed and discarded (NYC, 2000; USEPA, 2001). The only recommended approach currently available for addressing severe mold contamination is physical removal of mold-damaged materials. In severe cases, clean-up and repair of mold-contaminated buildings may be conducted using methods similar to those used for abatement of other hazardous substances such as asbestos (Shaughnessy and Morey, 1999). For example, in situations of extensive colonization (large surface areas greater than 100 ft² or where the material is severely degraded), extreme precautions may be required, including: full containment (complete isolation of work area) with critical barriers (airlock and decontamination room) and negative pressurization, personnel trained to handle hazardous wastes, and the use of full-face

respirators with HEPA filters, eye protection, and disposable full-body covering (NYC, 2000; ACGIH, 1999).

Physical removal interventions have proven effective, although additional research is needed regarding the containment of mold spores during the renovation process (NAS, 2000). In addition to strategies presented in the specific guidance documents listed above, an overview of the various recommended practices for the remediation of mold-contaminated materials, including porous, semi-porous and non-porous material removal, HVAC system remediation, containment strategies, and judging remediation effectiveness is presented in *Bioaerosols: Assessment and Control* (ACGIH, 1999; see Chapter 15, "Remediation of Microbial Contamination" by Shaughnessy and Morey).

The effect of biocides (to kill existing growth) and antimicrobials (to suppress or prevent growth) on mold varies according to mold species, and more research is needed to fully assess efficacy (NAS, 2000; Foarde, 1998; Cole and Foarde, 1999). The different chemical classes of biocides include alcohols, aldehydes, halogens, hydrogen peroxide, phenolics, and quaternary ammonium compounds (Foarde, 1998). In general, the use of biocides is discouraged by most experts because little research has been conducted on their effectiveness for this use and because of the potential human health hazards associated with this use (USEPA, 1997; Foarde, 1998; Cole and Foarde, 1999). In addition, research indicates that dead mold material often still retains the allergenic or toxic properties of the mold (Foarde, 1998; NAS, 2000), and thus replacement is often cited as the best mitigation option.

There are no registered biocides for treatment of porous duct materials (e.g., insulation) (Foarde, 1998), and mechanical cleaning has also been shown to be relatively ineffective. For example, a study investigating the effectiveness of mechanical cleaning of fibrous duct material contaminated with mold growth by vacuuming concluded that mechanical cleaning was only able to temporarily (for 6 weeks) reduce the surface mold load (Foarde et al., 1997b). Because of their potential to rapidly spread molds throughout a building, ventilation systems are of particular concern as mold contamination sources (Foarde et al., 1997b); USEPA guidelines recommend replacement (Foarde, 1998).

5.0 CURRENT RESEARCH AND INFORMATION GAPS

Possible areas of consideration for future research include:

Methodological Issues

- Standard methods for mold sampling
- Standard methods for analysis of mold toxins
- Standardized methods for analysis of mold allergens
- Determination of performance criteria for analytic methods (accuracy, detection limits, etc.)
- Information on factors that affect exposure and methods to quantify exposure from environmental samples (e.g., relationship between vacuum dust, etc. samples and actual exposure)

 Further research on fungal measurement using indicators of fungal growth (e.g., microbial VOCs)

Health Issues

- Health-based exposure standards or guidelines for mold
- Identification of threshold levels for sensitization to major residential mold allergens and for asthma exacerbation
- Additional data on standard amounts and types of molds (airborne and surface) in residential environments (with and without moisture problems) for comparison studies
- Appropriate levels of protection for mitigation workers

Building and Structural Issues

- Health impacts of building design and management
- Data to quantify which aspects of household water damage are related to respiratory illness
- Standard criteria for assessing water damage
- Standard, cost effective remediation procedures and criteria
- Effective and standard preventive measures

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Appendix A. Additional Internet Resources

In addition to the references and links appearing in the reference list above, the following table provides selected links with additional information on mold and mold contamination issues in homes.

| Sponsoring Organization/Topic | Internet Web Site Address | |
|---|--|--|
| Aerotech Laboratories, Inc Indoor air quality testing information | http://www.aerotechlabs.com/ | |
| Allergy, Asthma & Immunology Online | http://www.allergy.mcg.edu/ | |
| American Academy of Allergy, Asthma and Immunology | http://www.aaaai.org/ | |
| American Conference of Governmental Industrial Hygienists (ACGIH) | http://www.acgih.org/home.htm | |
| American Indoor Air Quality Council | http://www.iagcouncil.org/ | |
| American Industrial Hygiene Association (AIHA) Environmental | | |
| Microbiology Testing and Proficiency external peer review programs | http://www.aiha.org/micro.html | |
| (EMPAT and EMLAP) | | |
| American Society of Heating, Refrigerating and Air-Conditioning | http://www.ashara.assal | |
| Engineers, Inc. | http://www.ashrae.org/ | |
| Assessment Guide for Building Owners (EPA and NIOSH) | http://www.cdc.gov/niosh/baqtoc.html | |
| Asthma and Allergy Foundation of America | http://www.aafa.org/ | |
| Boston Medical Center Doc4Kids Program | http://www.bmc.org/program/doc4kids/index.html | |
| Canada Mortgage and Housing Corporation Healthy Housing & | http://www.cmhc-schl.gc.ca/cmhc.html (http://www.cmhc- | |
| Sustainability Projects | schl.gc.ca/en/imquaf/hehosu/index.cfm) | |
| Canada Mortgage and Housing Corporation Publications on moisture | http://www.cmhc-schl.gc.ca/en/imquaf/hehosu/hehosu_002.cfm | |
| and mold | http://www.chino-schi.gc.ca/en/inqua/nenosu/nenosu_ooz.chin | |
| Case Western Reserve University / General Clinical Research Center | http://gcrc.cwru.edu/stachy/default.htm | |
| Pulmonary Hemorrhage and Hemosiderosis In Infants | <u> </u> | |
| Center's for Disease Control and Prevention (CDC) | http://www.cdc.gov/ | |
| CDC – Air Pollution and Respiratory Health Branch | http://www.cdc.gov/nceh/asthma/default.htm | |
| CDC Questions and Answers on Stachybotrys chartarum and other | http://www.cdc.gov/nceh/asthma/factsheets/molds/default.htm | |
| molds | | |
| CDC Report on Cleveland Pulmonary Hemosiderosis and | http://www.cdc.gov/epo/mmwr/preview/mmwrhtml/mm4909a3.ht | |
| Stachybotrys | <u>m</u> | |
| Environmental Health Watch | http://www.ehw.org/ | |
| Environmental Microbiology and Mycology information from P&K | http://www.envirocenter.com/ | |
| Microbiology Services Inc. | | |
| Environmental Microbiology Laboratory, Inc. | http://www.emlab.com/ | |
| Health House Project of the American Lung Association | http://www.healthhouse.org/ | |
| Healthy Homes Partnership - USDA and HUD | http://www.uwex.edu/healthyhome/ | |
| HUD's Healthy Homes for Healthy Children | http://www.hud.gov/consumer/hhhchild.cfm | |
| HUD's Office of Healthy Homes and Lead Hazard Control | http://www.hud.gov/offices/lead/ | |
| Indoor Air Pollution: An Introduction for Health Professionals (USEPA) | http://www.epa.gov/iedweb00/pubs/hpguide.html | |
| Indoor Biotechnologies, ltd. | http://www.inbio.com/ | |
| Institute of Inspection Cleaning & Restoration fire and flood restoration | http://www.iicrc.org/ | |
| Johns Hopkins Asthma & Allergy | http://www.hopkins-allergy.org/ | |
| Master Home Environmentalist | http://www.alaw.org/air_quality/information_and_referral/master_ | |
| | home environmentalist/ | |
| Minnesota Department of Public Health - Mold in Homes | http://www.health.state.mn.us/divs/eh/aialr/iair/moldfs.html | |
| National Safety Council Indoor Air Program | http://www.nsc.org/ehc/indoor/iaq.htm | |
| New York City Department of Health Guidelines on Assessment and | http://nycdoitt.ci.nyc.ny.us/html/doh/html/epi/moldrpt1.html | |
| Remediation of Fungi in Indoor Environments | | |
| NIH National Institute of Environmental Health Sciences Asthma | http://www.niehs.nih.gov/airborne/home.htm | |
| Homepage | | |
| North Carolina State University Extension Service Mold, dust mites, | http://www.ces.ncsu.edu/depts/fcs/housing/docs/fcs3605.html | |
| fungi, spores, and pollen: Bioaerosols in the human environment | | |

| Safer Child, Inc. – Indoor Air Pollution | http://www.saferchild.org/indoor.htm |
|--|--|
| University of California Indoor Air Quality Tools: Education, Prevention and Investigation | http://ehs.ucsc.edu/ih/IAQC/IAQC-intro.html |
| University of Minnesota, Department of Environmental Health and Safety Fungi in Buildings | http://www.dehs.umn.edu/iaq/fungus/ |
| University of Montana Healthy Indoor Air | http://www.montana.edu/wwwcxair/ |
| USEPA Indoor Air Quality Homepage | http://www.epa.gov/iaq/ |
| USEPA Mold Resources | http://www.epa.gov/iaq/pubs/moldresources.html |
| USEPA Office of Children's Health Protection | http://www.epa.gov/children/ |
| USEPA Mold Remediation in Schools and Commercial Buildings | http://www.epa.gov/iaq/molds/index.html |